

WHAT IS CLAIMED IS:

1. A mutant form of 2,5-DKG reductase A having improved ability to convert 2,5-DKG into 2-KLG.

2. The mutant according to claim 1 having an amino acid substitution in a position of 2,5-DKG reductase A selected from the group consisting of 165, 166, 167, 168, 187, 188, 189, 190, 191, 192, 193, 194, 195, 196, 197, 198, 224, 225, 226, 227, 228, 229, 230, 231, 232, 233, 234, 262, 263, 264, 265, 266, 267, 268, 269, 270, 271, 272, 273, 274, 275, 276, 277, and 278.

3. The mutant according to claim 1 having an amino acid substitution in position 192.

4. The mutant according to claim 3 having an arginine at position 192.

5. A DNA construct comprising a structural gene containing at least one mutated codon, said gene coding for a mutant form of 2,5-DKG reductase A having improved ability to convert 2,5-DKG into 2-KLG.

6. A DNA construct according to claim 5, wherein said mutated codon is a codon selected from the group consisting of 165, 166, 167, 168, 187, 188, 189, 190, 191, 192, 193, 194, 195, 196, 197, 198, 224, 225, 226, 227, 228, 229, 230, 231, 232, 233, 234, 262, 263, 264, 265, 266, 267, 268, 269, 270, 271, 272, 273, 274, 275, 276, 277, and 278 which results in an amino acid substitution in a position of 2,5-DKG reductase A selected from the group consisting of 165, 166, 167, 168, 187, 188, 189, 190, 191, 192, 193, 194, 195, 196, 197, 198, 224, 225, 226, 227, 228, 229, 230, 231,

232, 233, 234, 262, 263, 264, 265, 266, 267, 268, 269, 270,
271, 272, 273, 274, 275, 276, 277, and 278.

7. A DNA construct according to claim 5, wherein said
mutated codon is codon 192 which results in an amino acid
substitution in position 192 of 2,5-DKG reductase A.

8. A DNA construct according to claim 7 which results
in arginine at position 192.

9. A mutant form of 2,5-DKG reductase A having
increased expression.

10. The mutant according to claim 9 having amino acid
substitutions in positions 2, 5, and 7 of 2,5-DKG reductase
A.

11. The mutant according to claim 10 having an
asparagine at position 2, a threonine at position 5, and a
serine at position 7.

12. A DNA construct comprising a structural gene
containing at least one nucleotide substitution, said gene
coding for a 2,5-DKG reductase A resulting in increased
expression of said 2,5-DKG reductase A.

13. A DNA construct according to claim 12, wherein
said nucleotide substitutions are in codons 2, 5, and 7
which result in amino acid substitutions in positions 2, 5,
and 7 of 2,5-DKG reductase A.

14. A DNA construct according to claim 13 which
results in asparagine at position 2, threonine at position
5, and serine at position 7.

15. A host cell transformed with an expression vector that includes a DNA construct according to claims 5, 6, 7, 8, 12, 13 or 14.

5 16. The host cell of claim 15 which is a bacterium.

17. The host cell of claim 16, wherein the bacterium is of the genus *Erwinia*.

10 18. The host cell of claim 16, wherein the bacterium is of the genus *Gluconobacter*.

15 19. The host cell of claim 16, wherein the bacterium is of the genus *Acetobacter*.

20 20. The host cell of claim 19, wherein the bacterium is *Acetobacter cerinus* (IFO 3263).

25 21. The host cell of claim 14, wherein the expression vector is a plasmid.

22. The host cell of claim 21, wherein the plasmid is pSStac.DKGR.AAA.HS1.

25 23. A mutant form of 2,5-DKG reductase A having improved temperature stability.

24. The mutant according to claim 23 having amino acid substitutions in positions 55 and 57 of 2,5-DKG reductase A.

30 25. The mutant according to claim 24 having an alanine at positions 55 and 57.

26. A DNA construct comprising a structural gene containing at least one mutated codon, said gene coding for a mutant form of 2,5-DKG reductase A having improved temperature stability.

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27. A DNA construct according to claim 26, wherein said mutated codons are codons 55 and 57 which result in amino acid substitutions in positions 55 and 57 of 2,5-DKG reductase A.

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28. A DNA construct according to claim 27 which results in alanine at positions 55 and 57.

29. A mutant form of 2,5-DKG reductase A having improved ability to convert 2,5-DKG into 2-KLG, having increased expression, and having improved temperature stability.

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30. The mutant according to claim 29 having amino acid substitutions in positions 192, 2, 5, 7, 55, and 57 of 2,5-DKG reductase A.

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31. The mutant according to claim 30 having an arginine at position 192, an asparagine at position 2, a threonine at position 5, a serine at position 7, and an alanine at positions 55 and 57.

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32. A DNA construct comprising a structural gene containing at least six mutated codons, said gene coding for a mutant form of 2,5-DKG reductase A having improved ability to convert 2,5-DKG into 2-KLG, having increased expression, and having improved temperature stability.

33. A DNA construct according to claim 32, wherein said codons are codons 192, 2, 5, 7, 55, and 57 which result in amino acid substitutions in positions 192, 2, 5, 7, 55, and 57 of 2,5-DKG reductase A.

34. A DNA construct according to claim 33 which results in arginine at position 192, asparagine at position 2, threonine at position 5, serine at position 7, and alanine at positions 55 and 57.

35. A host cell transformed with an expression vector that includes a DNA construct according to claims 32, 33 or 34.

36. The host cell of claim 35 which is a bacterium.

37. The host cell of claim 36, wherein the bacterium is of the genus *Erwinia*.

38. The host cell of claim 36, wherein the bacterium is of the genus *Gluconobacter*.

39. The host cell of claim 36, wherein the bacterium is of the genus *Acetobacter*.

40. The host cell of claim 39, wherein the bacterium is *Acetobacter cerinus* (IFO 3263).

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